FINAL REPORT

Microbiological Sampling Report

for

National Oceanic & Atmospheric Administration

Samplings Conducted on the Eleventh Floor
of Building SSMC-3
on February 17, 2000

Interagency Agreement #: D8H00CO31200
Task: 9903

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Prepared by

US Public Health Service

Division of Federal Occupational Health

Bethesda Central Office

Executive Summary

At the request of the National Oceanic & Atmospheric Administration (NOAA), Federal Occupational Health (FOH) conducted a microbiological sampling in room 11723 of Building SSMC-3, located at 1315 East-West Highway, Silver Spring, Maryland. Sampling was conducted on February 17, 2000. Air (both Andersenâ and Zefonâ), swab, contact plate, and vacuum dust samples were collected from this room and an indoor reference room 11839. Air samples were also collected from outdoors.

Findings are as follows:

· Indoor airborne fungal levels, by Andersen sampling, and spore levels, by Zefon, were lower than those of outdoors.

· In general, fungal burden on vertical surfaces was lower than that of horizontal surfaces.

· Fungal burden on horizontal surfaces of room 11839 was higher than that of room 11723. *Penicillium* was the predominant fungi recovered from contact plate samples.

· *Stachybotrys chartarum* was not detected from any air, swab, and contact plate samples collected.

· Very low fungal burden was detected from swab samples collected from surfaces of supply diffusers and return troughers in light fixture.

· The fungal level in plenum dust of room 11723 was at 10^3 CFU/g of fine dust level, a much lower level than that of the reference room 11839 (10^5 CFU/g of fine dust). *Stachybotrys chartarum* was detected from both samples. *Penicillium* and *Aspergillus* dominated the sample collected from room 11839.

· Fungal levels in carpet and furniture dust of room 11839 were at 10^3 - 10^4 CFU/g levels. Diverse fungal genera were detected from furniture dust while *Penicillium* and *Aspergillus* dominated the carpet dust sample. *Stachybotrys chartarum* was detected from both samples.

INTRODUCTION

At the request of the National Oceanic & Atmospheric Administration (NOAA), Federal Occupational Health (FOH) conducted a microbiological sampling in room 11723 of Building SSMC-3, located at 1315 East-West Highway, Silver Spring, Maryland. Sampling was conducted on February 17, 2000. Air (both Andersenâ and Zefonâ), swab, contact plate, and vacuum dust samples were collected from this room and an indoor reference room 11839. Air samples were also collected from outdoors.

EVALUATION METHODOLOGY

Air Samples

Various types of samples were collected from these rooms on February 17, 2000. Two types of air samples were collected from each room: (1) culturable method using Andersenâ N-6 samplers at a flow rate of 28.3 L/min, and (2) non-culturable method using Zefonâ Air-O-Cell cassettes at a flow rate of 15 L/min. Indoor Andersenâ air samples were collected for 3 minutes and outdoor samples were collected for both one and three minutes. Two percent (2 %) malt extract agar (MEA) and cellulose Czapek agar (CCA) was used to recover general fungi and cellulose-loving fungi, respectively. Non-culturable air samples were collected at the aforementioned sampling locations. Indoor samples were collected for ten minutes and outdoor samples were collected for both five and ten minutes. Outdoor air samples were collected near the entrance of the building. Temperature and relative humidity measurements were collected from each air sampling location by a battery operated, direct readout Hygroskopâ meter.

Contact Plate Samples

To determine fungal burden on horizontal and vertical surfaces of these rooms, four to eight contact plate samples were collected from each room. Samples were collected from randomly selected horizontal and vertical surfaces. Sampling was conducted by pressing the MEA-filled Rodacâ plate against the surface of interest for five seconds. A total of 12 contact plate samples were collected.

Swab Samples

Swab samples were collected from surfaces of each supply diffusers and return troughers in each room. They were collected by wiping a known area of surface with a sterile cotton swab (Culturette®) wetted with holding media. Approximately 5 in^2 area was wiped for return trougher and 4 in^2 for supply diffusers. The swab was then placed directly into its holder. Each holder was labeled with an identifiable number. A total of 8 swab samples were collected from these rooms.

Vacuum Dust Samples

Dust accumulated on carpeting, chairs and fabric system furniture, and the plenum were collected with a High Efficiency Particulate Air (HEPA) vacuum attached with a special "sock" device. For each carpet sample, a 3-ft by 3-ft area was vacuumed for at least five minutes. Total surface areas of 9 ft^2 were vacuumed from system furniture and chairs, and composite as one sample. Dust accumulated above the ceiling plenum was also vacuumed and composite as one sample. One plenum dust sample was collected from each room. One carpet and one composite furniture dust samples were collected from the reference room 11839.

All samples collected were sent for next morning delivery to FOH's Environmental Microbiology Laboratory (EML) in Philadelphia, Pennsylvania for analysis.

Laboratory Procedures

Upon receipt, all Andersen® air and contact plate samples were incubated in a 25°C incubator. Each swab sample was suspended in sterile distilled water, diluted serially, and inoculated onto agar plates. Both MEA and CCA were used for retrieving fungi. At least three dilution series were used for each sample. Each vacuum dust sample was sieved through a 250 mm sieve. The fine dust (< 250 mm) retrieved was then weighed and followed the dilution plating for fungal analysis.

All plates were incubated in a 25°C incubator. They were examined every other day for up to 10 days to ensure the full recovery of fungi. Fungal identification was based on colony morphology, spores and conidia formation. Total fungal colonies formed on each MEA plate and *Stachybotrys chartarum* on CCA plates were counted and recorded. Fungal levels in samples were presented as colony forming units (CFUs) per measuring unit. For example, CFU/m^3 for Andersen® air samples, CFU/in^2 for wipe samples, CFU/plate for contact plate samples, and CFU/g of fine dust for vacuum dust samples.

All Zefon® cassette samples were analyzed by the Environmental Microbiology Laboratory in Escondido, California for direct microscopic examination. Fungal spores were identified and their airborne levels were presented as spores/m^3.

RESULTS AND DISCUSSION

Temperature and Relative Humidity

Indoor temperature and relative humidity measurements ranged from 73.8°F to 74.6°F, and 16.5% – 17.5%, respectively (Table 1). Outdoors temperature reading was lower (47.5 °F) , but with a slightly higher relative humidity (20.9%).

Microbiological Analyses Results

All laboratory analytical reports from FOH's EML are presented in Attachment A in a laboratory report #NOAA-00-33R. Results from microscopic examination of Zefonâ cassette samples are presented in Attachment B.

Air Samples

Andersen Results

Indoor airborne fungal levels were lower than those of outdoors (Table 1). *Cladosporium* dominated outdoor air samples while *Penicillium* was the predominant indoor fungi. *Stachybotrys chartarum* was not detected from these samples.

Zefon Results

Outdoor fungal spores levels were higher than those of indoors (Table 1). Predominant spore types detected from outdoors were *Cladosporium* and Basidiospores. Besides those spore types detected outdoors, *Penicillium/Aspergillus* types were also detected in room 11839. *Stachybotrys chartarum* was not detected from any sample collected.

Table 1. Temperature and relative humidity measurements and airborne fungal levels at different rooms of the 11th floor in SSMC-3 on February 17, 2000.

Rooms Parameters	11839 Reference#	11723	Outdoors
Temperature (°F)	74.6	73.8	47.5
Relative Humidity (%)	17.5	16.5	20.9
Airborne Fungal Levels (CFU/m^3)	130	165	200* 212
Total Fungal Spores ($Spores/m^3$)	107	7	294* 280

Indoor reference.

* Two samples were collected from outdoors.

Wipe Samples

Most (7 out of 8) samples collected from surfaces of supply diffusers and return troughers in light fixtures were below the detection limits (BDL) (3 CFU/in^2 for supply diffuser and 2 CFU/in^2 for return trougher). The only sample showing fungal growth was collected from supply diffuser surfaces in room 11839 with 3 CFU/in^2 of *Chaetomium*.

Contact Plate Samples

In general, higher fungal levels were detected from the horizontal surfaces than vertical surfaces (Table 2). Fungal levels on vertical surfaces ranged from BDL of 1 CFU/plate to 2 CFU/plate. Fungal levels on horizontal surfaces ranged from 1 CFU/plate to 36 CFU/plate. Higher fungal levels on horizontal surfaces were detected from the reference room 11839. *Penicillium* was the predominant fungal genus recovered from room 11839.

Vacuum Dust Samples

Reference Room 11839

Samples were collected from plenum, carpet, and furniture. Fungal levels in these fine dust samples were at the levels of $10^3 – 10^5$ CFU/g of fine dust (Table 3). A diverse fungal population was detected from furniture dust with *Cladosporium* and *Alternaria* as the predominant fungi. However, *Penicillium* and *Aspergillus* dominated carpet and plenum dust samples collected from this room. *Stachybotrys chartarum*

was detected on every sample collected from this room (Table 3).

Room 11723

The only sample collected from this room was plenum dust. Fungal level was at 10^3 CFU/g of fine dust, lower than that of the reference room 11839. *Penicillium* was the predominant fungal genus recovered followed by *Aureobasidium* and *Paecilomyces. Stachybotrys chartarum* was detected from this sample.

Table 2. Fungal levels (CFU/plate) on horizontal and vertical surfaces of different rooms at the 11th floor of SSMC-3, by contact plate sampling collected on February 17, 2000.

Rooms	**11839** **Reference#**	**11723**
Horizontal Surfaces (CFU/plate)	11 – 36* (4**)	1 - 2 (3)
Vertical Surfaces **(CFU/plate)**	<1 - 2 (4)	<1 (4)

Indoor reference.

* Ranges.

** Total number of samples.

Table 3. Total fungal levels (CFU/g of fine dust) in fine dust collected from carpet, plenum, and furniture of rooms 11723 and 11839 of SSMC-3, by vacuum dust sampling, collected on February 17, 2000.

Rooms	**11839** **Reference#**	**11723**
Plenum **(CFU/g of fine dust)**	144,000 (+*)	3,168 (+)
Carpet **(CFU/g of fine dust)**	28,800 (+)	NA**
Furniture **(CFU/g of fine dust)**	8,119 (+)	NA

Indoor reference.

*+: *Stachybotrys chartarum* was detected on MEA and/or CCA plates.

-: *Stachybotrys chartarum* was not detected on MEA and CCA plates.

** Not applicable.

CONCLUSIONS

· Indoor airborne fungal levels, by Andersen sampling, and spore levels, by Zefon, were lower than those of outdoors.

· In general, fungal burden on vertical surfaces was lower than that of horizontal surfaces.

· Fungal burden on horizontal surfaces of room 11839 was higher than that of room 11723. *Penicillium* was the predominant fungi recovered from contact plate samples.

· *Stachybotrys chartarum* was not detected from any air, wipe, and contact plate samples collected.

· Very low fungal burden was detected from wipe samples collected from surfaces of supply diffusers and return troughers in light fixture.

· The fungal level in plenum dust of room 11723 was at 10^3 CFU/g of fine dust level, a much lower level than that of the reference room 11839 (10^5 CFU/g of fine dust). *Stachybotrys chartarum* was detected from both samples. *Penicillium* and *Aspergillus* dominated the sample collected from room 11839.

· Fungal levels in carpet and furniture dust of room 11839 were at 10^3 - 10^4 CFU/g levels. Diverse fungal genera were detected from furniture dust while *Penicillium* and *Aspergillus* dominated the carpet dust sample. *Stachybotrys chartarum* was detected from both samples.

RECOMMENDATIONS

· Conduct thorough cleaning of these rooms by HEPA vacuuming of porous materials and wet-wiping of hard surfaces.

· Conduct any above ceiling plenum work after hour. Thoroughly HEPA vacuum the surrounding areas afterwards.

· Implement an emergency water intrusion protocol for this building to adequately manage any unexpected water intrusion in order to prevent fungal proliferation.

ATTACHMENT A

Microbiological laboratory report #NOAA-00-33R for samples collected from eleventh floor of SSMC-3, on February 17, 2000.

ATTACHMENT B

Results from microscopic examination of Zefon air samples collected from eleventh floor of SSMC-3, on February 17, 2000.

USPHS DFOH ENVIRONMENTAL MICROBIOLOGY LABORATORY, PHILADELPHIA, PA

LABORATORY REPORT #NOAA-00-33R

Client agency: National Oceanic and Atmospheric Administration, Silver Spring, MD

POIS#/task #: D8H00CO31200 / 9903

Sampling date: 2/17/00

Dates of inoculation: 2/17/00 (airs and contact plates), 2/18/00 (wipes), and 2/20/00 (dust)

General location: SSMC-3, Silver Spring, MD

Specific location: 11th floor

Sampling techniques: Air (Andersen N-6 sampler), contact plate, wipe, and vacuum dust samplings

Medium used: Malt extract agar (MEA) and cellulose Czapek agar (CCA) for fungi

Samples submitted by: J. Sobelman

Date characterization completed: 3/1/00

(A) Air samples on MEA and CCA plates

Sample ID	Sampling Location	Air Volume (L)	Fungi on MEA @ 25º C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
3-11839-0217A1, A2	11th floor, room 11839, office, control	84.9	1. *Penicillium* (11*) CFU/m^3 = 130	No
3-11723-0217A1, A2	11th floor, room 11723, center of storage room	84.9	1. *Penicillium* (13) 2. *Cladosporium* (1) CFU/m^3 = 165	No
3-OA1-0217, 3-OA2-0217	Outside bldg. 3	84.9	1. *Cladosporium* (13) 2. *Penicillium* (2) 3. *Epicoccum* (1) 4. *Paecilomyces* (1) CFU/m^3 = 200	No
3-OA1-0217, 3-OA2-0217	Outside bldg. 3	28.3	1. *Cladosporium* (5) 2. Basidiomycetes (1) CFU/m^3 = 212	No
FB	Field blank	NA#	No fungal growth	No
SB	Shipping blank	NA	No fungal growth	No

(B) Contact plate samples on MEA plates

Sample ID	Sampling Location	Fungi detected on MEA @ 25º C
3-11839-0217CP1	11th floor, room 11839, column between windows	**No fungal growth** CFU/plate < 1
3-11839-0217CP2	11th floor, room 11839, wall above light switch	**1. yeast (1)** CFU/plate = 1
3-11839-0217CP3	11th floor, room 11839, top of desk	**1. *Penicillium* (28)** 2. *Cladosporium* (4) 3. *Alternaria* (2) 4. *Paecilomyces* (2) **CFU/plate = 36**
3-11839-0217CP4	11th floor, room 11839, top of printer	**1. *Penicillium* (11)** **CFU/plate = 11**
3-11839-0217CP5	11th floor, room 11839, top of grey lateral file	**1. *Penicillium* (20)** 2. *Alternaria* (3) 3. *Aureobasidium* (1) 4. *Bipolaris* (1) 5. *Cladosporium* (1) 6. *Paecilomyces* (1) **CFU/plate = 27**
3-11839-0217CP6	11th floor, room 11839, top of computer	**1. *Penicillium* (7)** 2. *Mucor* (1) 3. *Paecilomyces* (1) 4. Basidiomycetes (2) **CFU/plate = 11**

3-11839-0217CP7	11th floor, room 11839, front of system furniture	**1. *Aspergillus niger*** (1)** 2. *Chaetomium* (1) **CFU/plate = 2**
3-11839-0217CP8	11th floor, room 11839, front of grey lateral file	**No fungal growth** CFU/plate < 1

Sample ID	Sampling Location	Fungi detected on MEA @ 25º C
3-11723-0217CP1	11th floor, room 11723, front of grey cabinet	**No fungal growth** CFU/plate < 1
3-11723-0217CP2	11th floor, room 11723, grey shelf with files	**1. *Trichoderma* (1) CFU/plate = 1**
3-11723-0217CP3	11th floor, room 11723, top of safe	**1. *Penicillium* (2) CFU/plate = 2**
3-11723-0217CP4	11th floor, room 11723, blue shelf near safe	**1. *Cladosporium* (1) CFU/plate = 1**

(C) Wipe samples on MEA and CCA plates

FOH ID	Sample ID	Sampling Location	Area (in^2)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
Blank	Blank	Blank	NA	10X-MEA 10X-CCA	No fungal growth	No
W011	3-11839-0217R1	11th floor, room 11839, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W12	3-11839-0217R2	11th floor, room 11839, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No

W13	3-11839-0217S1	11th floor, room 11839, supply	4	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 3	No
W14	3-11839-0217S2	11th floor, room 11839, supply	4	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 3	No
W15	3-11839-0217S3	11th floor, room 11839, supply	4	10X-MEA 10X-CCA	1. *Chaetomium* (1) CFU/in^2 = 3	No

FOH ID	Sample ID	Sampling Location	Area (in^2)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
W16	3-11723-0217R1	11th floor, room 11723, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W17	3-11723-0217R2	11th floor, room 11723, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W18	3-11723-0217S1	11th floor, room 11723, supply	4	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 3	No

(D) Vacuum dust samples on MEA and CCA plates

FOH ID	Sample ID	Sampling Location	Weight (g)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
V01	3-11723-0217V01	11th floor, room 11723, above ceiling	0.101	40X-MEA 10X-CCA	1. *Penicillium* (4) 2. *Aureobasidium* (2) 3. *Paecilomyces* (2) **CFU/g = 3,168**	Yes (1) CFU/g = 99

FOH ID	Sample ID	Sampling Location	Weight (g)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25° C
V02	3-11839-0217V01	11th floor, room 11839, furniture	0.101##	40X-MEA 10X-CCA	**1. *Cladosporium* (11)** 2. *Alternaria* (8) 3. *Epicoccum* (6) 4. *Penicillium* (5) 5. *Aureobasidium* (4) 6. *Aspergillus sp.* (2) 7. *Bipolaris* (2) 8. *Aspergillus niger*** (1) 9. *Chaetomium* (1) 10. *Trichoderma* (1) CFU/g = 8,119	Yes (1) CFU/g = 50
V03	3-11839-0217V02	11th floor, room 11839, carpet	0.100	40X-MEA 10X-CCA	**1. *Penicillium* (13)** 2. *Aspergillus niger*** (6) 3. *Alternaria* (3) 4. *Aureobasidium* (3) 5. *Cladosporium* (3) 6. yeast (44) CFU/g = 2.9 x 10^4	Yes (2) CFU/g = 200

FOH ID	Sample ID	Sampling Location	Weight (g)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
V04	3-11839-0217V03	11th floor, room 11839, above ceiling	0.100	400X-MEA 10X-CCA	1. *Penicillium* (21) 2. *Aspergillus niger*** (8) 3. *Aspergillus sp.* (2) 4. *Cladosporium* (2) **5. *Aspergillus flavus**** (1)** 6. *Chaetomium* (1) 7. *Trichoderma* (1) CFU/g = 1.4×10^5	Yes (4) CFU/g = 400

* Colony counts.

** Opportunistic fungi.

*** Toxigenic fungi.

Not applicable.

5ml of sterilized distilled water were added instead of 10ml.